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Lymphoma

Primary Vitreoretinal Lymphoma: A Report from an International Primary Central Nervous System Lymphoma Collaborative Group Symposium

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ABSTRACT

Primary vitreoretinal lymphoma (PVRL), also known as primary intraocular lymphoma, is a rare malignancy typically classified as a diffuse large B-cell lymphoma and most frequently develops in elderly populations. PVRL commonly masquerades as posterior uveitis and has a unique tropism for the retina and central nervous system (CNS). Over 15% of primary CNS lymphoma patients develop intraocular lymphoma, usually occurring in the retina and/or vitreous. Conversely, 65%–90% of PVRL patients develop CNS lymphoma. Consequently, PVRL is often fatal because of ultimate CNS association. Current PVRL animal models are limited and require further development. Typical clinical findings include vitreous cellular infiltration (lymphoma and inflammatory cells) and subretinal tumor infiltration as determined using dilated fundoscopy, fluorescent angiography, and optical coherent tomography. Currently, PVRL is most often diagnosed using both histology to identify lymphoma cells in the vitreous or retina and immunohistochemistry to indicate monoclonality. Additional adjuncts in diagnosing PVRL exist, including elevation of interleukin-10 levels in ocular fluids and detection of Ig_H or T-cell receptor gene rearrangements in malignant cells. The optimal therapy for PVRL is not defined and requires the combined effort of oncologists and ophthalmologists. PVRL is sensitive to radiation therapy and exhibits high responsive-

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ness to intravitreal methotrexate or rituximab. Although systemic chemotherapy alone can result in high response rates in patients with PVRL, there is a high relapse rate. Because of the disease rarity, international, multicenter, collaborative efforts are required to better understand the biology and pathogenesis of PVRL as well as to define both diagnostic markers and optimal therapies. ***The Oncologist*** 2011;16:1589–1599

Introduction

The most common lymphoma of the eye is primary vitreoretinal lymphoma (PVRL), also known as "primary intraocular lymphoma," a rare subset of primary central nervous system lymphoma (PCNSL) [1]. Approximately 15%–25% of patients with PCNSL have or ultimately develop an ocular manifestation of their lymphoma. Conversely, 56%–90% of patients with PVRL consequently have or eventually develop CNS disease. Because of its rarity, PVRL is difficult to study, and no clear standards exist for diagnosis, monitoring, and therapy. PVRL is still a challenging malignancy with a high mortality rate and significant morbidity. The Fifth Annual, National Cancer Institute–sponsored International PCNSL Collaborative Group (IPCG) conference, a multidisciplinary meeting, conducted a symposium on PVRL. The symposium is summarized herein, including sections on tumor biology, nomenclature, epidemiology and prognosis, biology and pathogenesis, animal models, clinical manifestations, diagnosis, therapeutics, and future investigations.

Nomenclature

Intraocular lymphomas represent a heterogeneous group of malignancies that are located in different tissues within the eye. Each of the intraocular lymphomas has different morphological, immunophenotypical, and genetic features, with completely different clinical courses [2, 3]. It is, therefore, preferable to refer to the various forms of intraocular lymphoma according to whether they are vitreoretinal, choroidal, ciliary, or iridal and whether they are primary or secondary to CNS lymphoma (CNSL) or disseminated, systemic disease. They are then subtyped histomorphologically according to the World Health Organization (WHO) Lymphoma Classification [2, 3].

The most common intraocular lymphoma is PVRL, which is a high-grade (i.e., aggressive) lymphoma, usually of the B-cell type. It can be subtyped as diffuse large B-cell lymphoma (DLBCL) [2, 3]. Rarely, PVRL of T-cell–rich B-cell lymphoma and the T-cell type have been described [4–6].

The second major group of intraocular lymphomas is the uveal lymphomas, which can be subdivided into primary neoplasms of the choroid, iris, and ciliary body as well as secondary choroidal lymphomas in patients with disseminated disease [2]. Primary choroidal lymphomas were first recognized by Triebenstein in 1920 [7], and at least 100 cases have since been described in the literature. In contrast to high-grade malignant PVRL, primary choroidal lymphomas are low-grade (i.e., indolent) B-cell lymphomas. They are typically extranodal marginal zone B-cell lymphomas (EMZLs), according to the WHO classification, similar to the EMZLs that more commonly occur in the ocular adnexa, for example, conjunctiva [8]. The primary choroidal lymphomas do not have any association with CNS disease. Because of its typically low-grade nature and indolent clinical course without retinal and vitreal involvement, primary choroidal lymphoma has been previously termed "uveal or intraocular pseudotumor" and "reactive lymphoid hyperplasia" [9]. However, several investigators have confirmed lymphoma monoclonality [8–11]. Primary iridal lymphomas are exceptionally rare, with fewer than a dozen cases reported in the literature [2]. Interestingly, B-cell and T-cell lymphomas arise in the iris in equal measure. Secondary choroidal lymphomas represent metastatic systemic lymphoma and are usually confined to the choroid [2]. Although secondary lymphomatous ocular disease with predominant involvement of the retina and without uveal infiltration has been reported, this is extremely rare. The most common systemic lymphoma subtype secondarily involving the choroid is DLBCL. This is followed by multiple myeloma, extramedullary plasmacytoma, lymphoplasmocytic lymphoma/immunocytoma (including Waldenström's macroglobulinemia), and B-cell chronic lymphocytic leukemia [2]. Rarely, secondary intraocular disease arises from intravascular lymphoma with secondary involvement of the eye [12, 13].

Epidemiology

The incidence of PVRL is difficult to estimate because no central database exists for this disease. The incidence of all eye and orbital malignancies was 0.8 per 100,000 persons in 2007, and these were more common in patients aged ≥50 years [14]. Retinoblastoma, choroidal melanoma, and orbital lymphoma probably account for the bulk of eye and orbital tumors, not intraocular lymphoma, which is considered rare.

Better data exist for PCNSL. The Central Brain Tumor Registry of the U.S. recorded an incidence of 0.46 (95% confidence interval [CI], 0.45–0.47) per 100,000 person-years in 2004–2007; the incidence was higher in males (0.54) than in females (0.39), with a male:female ratio of 1.38, in contrast to the female predominance usually reported for intraocular lymphoma [15]. There was no statistical difference in the proportion of brain tumors attributed to lymphoma among races. The age at peak incidence was 75–84 years, with an incidence of 2.08 (95% CI, 2.06–2.32) per 100,000 person-years [16]. Although there have been concerns that the incidence of PCNSL is increasing, equivalent data from 1990–1994 reported by Surawicz showed a similar overall incidence of 0.43 ± 0.2 per 100,000 person-years, with a mean age at diagnosis of 54 years [17]. The earlier higher number of cases of PCNSL may partially have been attributable to AIDS-related immunodeficiency, a factor that is likely of less importance in the era of highly active antiretroviral therapy [18].

Knowledge of the incidence of PCNSL is useful, because it

is estimated from multiple case series that intraocular involvement occurs in 15%–25% of cases [19–21]. Conversely, 65%–90% of patients presenting with PVRL develop CNSL, usually within 29 months [15]. Therefore, of the estimated 1,927 incident cases of PCNSL in 2006, ~20%, or 380 cases, would have had ocular involvement. PVRL patients who not develop intracranial involvement probably account for no more than an additional 50 cases per year. This estimate of roughly 380 incident cases of PVRL in the U.S. annually is consistent with that of Chan and Gonzales [1] and with the ~21 incident cases per 100,000 patients with ocular disorders presenting at referral eye centers per year in Japan [22].

The visual prognosis for patients with PVRL is not clearly defined in the literature as a result of factors that include: the small number and small size of published series, the absence of visual acuity reporting in some series, and variable follow-up and treatment modalities within and between series. In one large series, Frenkel et al. [23] reported wide variation in presenting visual acuities, from normal to hand motion. They further noted that, in general, visual acuity did not change substantially with treatment, although dramatic improvements in poor visual acuity were possible if treatment was rapidly instituted following the onset of clinical ocular involvement. A previous publication from the IPCG addressed CNS progression and mortality of PVRL patients in 83 HIV^- individuals with PVRL and without clinical or radiological evidence of CNS involvement who received a wide spectrum of treatments [24, 25]. The median overall survival (OS) time, which was not impacted by the therapeutic approach, was 58 months. At final follow-up (median for surviving patients, 32 months), progression to brain involvement had occurred in 34.9% of patients, and 57.6% of 33 patients had died as a result of CNSL.

Biology and Pathogenesis of PVRL and PCNSL

Biological studies of PVRL are limited because of a paucity of tissue specimens and the rarity of this condition. Nevertheless, it is likely that insight into the biology and pathogenesis of PVRL can be realized from the more extensive studies that have been conducted in PCNSL patients. Three major properties of PCNSL/PVRL biology are apparent: (a) throughout their natural history, it is extremely rare for PCNSLs and PVRLs to manifest or recur outside the brain, demonstrating a unique tropism for the CNS; (b) PCNSL and PVRL are associated with a prognosis that is inferior to that of other localized extranodal subtypes of non-Hodgkin's lymphoma tumors confined to a single extranodal site, such as the bone; and (c) PCNSL exhibits a high responsiveness to methotrexate, with concomitant lower responsiveness to doxorubicin-based chemotherapy regimens [26]. The molecular basis for this differential sensitivity to chemotherapy has not been deduced, but the impact of methotrexate is significant in that ~20% of CNSL patients have long progression-free survival (PFS) intervals with methotrexate-based monotherapy [27].

The origin of the tumor cells in PCNSL and PVRL is unknown. One possibility is that a malignant clone of B cells of systemic origin might evolve to express selective, specific adhesion molecules that facilitate homing to the meninges, brain parenchyma, and intraocular compartments (retina, vitreous, and optic nerve), whereby secondary mutations facilitate further growth. Recent studies have demonstrated that tumor clones related to CNSL can be detected in the blood and bone marrow of PCNSL patients, suggesting that the CNS and PVRL microenvironment might favor tumor progression [28, 29]. In immunosuppressed populations, PCNSL is typically associated with the infection of neoplastic B cells by the Epstein-Barr virus (EBV) [30]. EBV infection of normal B cells may result in their immortalization; however, proliferation of EBV-infected B cells is held in check by T-cell immunity. With diminished T-cell function, however, EBV-infected lymphocytes may evolve to malignancy [31].

Chemokines regulate leukocyte trafficking, proliferation, and adhesion. Chemokine (C-X-C motif) ligand 13 (CXCL13), a B-cell attracting chemokine, has been identified in PCNSL tumors [32]. CXCL13 is a lymphoid chemokine that may promote B-cell homing to secondary lymphoid organs [33]. Expression of the chemokine stromal derived factor-1 (CXCL12) has also been associated with malignant B cells in PCNSL and PVRL [34, 35]. It has been hypothesized that the ectopic expression of these chemokines within the intraocular compartment may contribute to lymphoma homing to the retinal pigment epithelium (RPE) from the choroidal circulation [36].

PCNSL tumors were recently analyzed in large-scale transcriptional profiling studies using microarrays [37, 38]. Upregulation of the proto-oncogenes c-*myc* and *Pim-1* as well as the ectopic expression of the B-cell growth factor interleukin (IL)-4 are found within PCNSLs. In addition, the activated form of the transcription factor signal transducer and activator of transcription (STAT)-6, a mediator of IL-4–dependent gene expression, was found in lymphoma cells and tumor endothelia in PCNSL, suggesting a significant functional role of IL-4 signaling in the pathogenesis of this type of lymphoma. The intratumoral expression of STAT-6 protein (encoded by an IL-4–induced gene) was associated with s short survival time in PCNSL patients treated with high-dose methotrexate [39]. These data support the hypothesis that the expression pattern of STAT-6 might constitute a novel biomarker for prognostic determination in PCNSL patients. The gene expression study of Tun et al. [38] compared PCNSL and non-CNSL and highlighted the differential expression of extracellular matrix molecules in PCNSL, in particular, osteopontin and chitinase 3-like-1 [37, 38].

The limitations of the aforementioned studies are that each study used distinct microarray platforms and analyzed a relatively small number of specimens, and each lacked an independent validation set of tumor samples. It is, therefore, noteworthy that a subset of overlapping genes that are concordant in distinguishing PCNSL from non-CNS large cell lymphoma was elucidated [40]. The majority of these are components of the extracellular matrix. For example, osteopontin and chitinase-3-like-1 were independently expressed at higher levels in PCNSL cases. In contrast, collagen type VI, laminin α-4, and lumican, a keratin sulfate proteoglycan, were

expressed at higher levels in systemic lymphoma in each study. In addition, each analysis detected greater expression of genes involved in signaling and cell proliferation. For example, regulator of G-protein signaling (RGS)-13 was expressed at twofold higher levels in PCNSLs than in systemic lymphomas. RGS proteins negatively regulate the signaling of G-proteins, including chemokine receptor 18 [41].

There is an accumulation of evidence that noncoding short microRNAs (miRNAs) promote or suppress cell transformation as well as regulate tumor invasion and metastasis [42]. Robertus et al. [43] demonstrated significantly higher expression of the miRNA miR-17–5p in CNSLs than in nodal and testicular DLBCL. miR-17–5p microRNA is believed to promote tumor growth via the downmodulation of negative regulators of mitogen-activated protein kinase signaling [44]. It will be interesting to determine the functional significance of the differential expression of miRNAs in terms of CNS tropism and the pathogenesis of lymphoma within the brain and ocular microenvironments.

Roy et al. [45] conducted an in-depth proteomic analysis of cerebrospinal fluid (CSF) from CNSL patients using two-dimensional liquid chromatography and mass spectrometry. As controls, CSF was obtained from patients with nonmalignant CNS conditions, including patients with systemic cancer. A high degree of concordance between the two sets of results was noted (Spearman correlation, 0.71). At least 80 differentially expressed CSF proteins were identified, both upregulated in CSF from CNSL patients and downregulated relative to CSF from control subjects [45]. The vast majority of peptides upregulated in CNSL were serine proteases or protease inhibitors, complement mediators and inhibitors, and components of the extracellular matrix, notably osteopontin and chitinase-3-like-1, as predicted by gene expression analyses [38, 40]. Most of the proteins downmodulated in CSF of CNSL patients were associated with normal neuronal function, including neuropeptides that regulate neuronal signaling. To validate this technology, the investigators defined the CSF expression of the serine protease inhibitor antithrombin III (ATIII) using a different technology, enzyme-linked immunosorbent assay (ELISA). ATIII was of particular interest because this peptide was independently identified by transcriptional profiling to be highly expressed in CNSL patients in cases associated with a short survival duration. Elevation of CSF ATIII in CNSL cases was confirmed by ELISA in an independent validation set of 100 cases. However, elevated ATIII in CSF was also noted in glioblastoma as well as metastatic carcinoma patients, highlighting the need to identify protein biomarkers that are specific to CNSL.

The aforementioned investigations in CNSL may offer insight into the pathogenesis of PVRL, although studies focused on the latter are necessary to determine the unique biological features of lymphomas affecting the eye.

Animal Models

Animal models are critical to better understand the pathophysiology of PVRL, for which no human cell line is currently available. They also provide an indispensable tool with which to evaluate new treatment strategies. The first two animal models of ocular lymphoma used murine lymphoma cells of T-cell lineage (Rev-2-T-6 cells) and were more models of murine metastatic malignant lymphomas rather than models of PVRL [46, 47]. These models used i.p. injection of Rev-2-T-6 cells into newborn mice to develop cerebral and ocular lesions. The first model of PVRL was developed by Chan et al. [48] after intravitreal injection of the same cell line into Balb/c mice. Despite the use of a lymphoma cell line of T-cell origin, this model provided the first evidence of the major role of the RPE as a barrier for the development of PVRL and confirmed the role of IL-10 in the diagnosis of PVRL by demonstrating high levels of IL-10 mRNA transcripts in the eyes of animals bearing a tumor. T-helper 1 (Th1) cytokines such as interferon-γ were also detected in the absence of IL-2. This latter finding was later confirmed in a model of B-cell PVRL [49], which is more relevant because most PVRLs, similar to PCNSLs, are B-cell lymphomas.

The mechanisms involved in the migration of tumor cells between the eye and brain constitute a major issue in the development of therapeutic strategies that could modify the natural history and prognosis of the disease. Chemokine receptor 1 (a T-cell chemokine receptor) transcripts were detected in the eyes of animals developing ocular T-cell tumors, suggesting a possible role for this chemokine in the homing of tumor cells [48, 50]. Similarly, C-X-C chemokine receptor (CXCR)-4 and CXCR-5 molecules were detected in a murine model of B-cell lymphoma [51]. This observation is of interest, because it points to those molecules as potential candidate markers for tumor cell migration between the eye and brain. CXCR-4 and CXCR-5 have also been identified in the eyes of three patients with PVRL [34]. Li et al. [51] developed a model of PVRL in severe combined immunodeficient (SCID) mice using human lymphoma B cells injected intravitreally. With this model, the authors evaluated a novel therapeutic strategy using a single intravitreal injection of the lymphotoxin HA22 to successfully treat intraocular lymphoma. Touitou et al. [49] developed the first model of B-cell PVRL in immunocompetent mice, using a syngeneic B-cell lymphoma. This work demonstrated the role not only of the tumor but also of the microenvironment in tumor pathogenesis. Escape from an immune-privileged site in this model included partial inhibition of a Th1/Tc1 cytokine profile of infiltrating T lymphocytes, the recruitment of natural regulatory T cells, and the generation of induced regulatory T cells, arising not only from the spleen but also from possible phenotypical conversion into the eye itself. The multitude of escape mechanisms developed either by the tumor or by its microenvironment provides potential therapeutic targets for future studies. Finally, Mineo et al. [52] developed a murine model of PVRL in SCID mice using a human lymphoma B cell line expressing the human surface molecule CD20 to demonstrate the efficacy of intravitreal injection of monoclonal antibodies directed against CD20.

Clinical Manifestations

Insidious onset and delay in diagnosis of PVRL are common. Patients may complain of vitreous floaters for 1–2 years before lymphoma is suspected. Patients are usually either told that the floaters are a result of normal degenerative changes or that they have uveitis. Injectable or oral corticosteroids may be prescribed, with temporary improvement. Death of lymphoma cells at the level of the RPE can be interpreted as an indication that anti-inflammatory treatment is working. Even if the diagnosis is suspected, the quantity of vitreous cells may be deemed too few to permit an adequate biopsy until further proliferation occurs.

The clinician who makes the diagnosis of PVRL is assisted by a variety of clinical observations [15, 22, 53]. Most cases have little anterior segment inflammation. The eye is white and quiet. Posterior synechiae are never present. Reactive T cells may contribute to iritis and even keratic precipitates, but usually the infiltrating cells are confined to the vitreous cavity. In the vitreous, a lymphomatous infiltrate typically produces sheets and clusters of cells. Cells are relatively homogeneous, may appear larger than ordinary inflammatory cells, and do not clump into reactive clusters, causing an aurora borealis appearance as the cells line up along the vitreous fibriles (Fig. 1). Vitreous haze is generally less than expected for the number of cells and may be oddly accentuated in a peripheral or superior location that does not reflect the gravitational effect seen in uveitis.

Retinal or RPE involvement is helpful when present. Some associated findings are nonspecific, such as retinal vascular leakage, and are not helpful in distinguishing lymphoma from uveitis. Cells can directly infiltrate the retina and grow focally creating a semiopaque, gray spot that appears homogenous on optical coherent tomography (OCT) (Fig. 2). More commonly, lymphoma cells grow along Bruch's membrane under the RPE. Because RPE cells are bound by tight junctions and can stretch, local growth can create focal, solid detachments of the RPE. The cells can continue to proliferate and spread under the retina in large sheets of yellow-white cells. Islands of pigment seem to float on these deposits confirming their sub-RPE origin. With early or diffuse involvement, so-called leopard-spot pigmentation occurs (Fig. 3); it is better appreciated on photographs enhanced with venous fluorescein angiography (Fig. 4) [54]. If the cells die, the pigmentary alterations persist.

Either retinal or RPE infiltration damages structures essential for vision and is an absolute indication for treatment when progressive in order to save vision. Vitreous cellular infiltration produces symptoms of blurred vision, but objectively the vision is often excellent unless there is direct lymphomatous infiltration of the central macula. Cystoid macular edema, which is commonly found in nonlymphomatous uveitis, is rare in PVRL.

Diagnosis

The diagnosis of PVRL requires the definitive identification of malignant lymphoid cells in the eye. Diagnostic procedures are often performed based on strong clinical suspicion [55]. A history of an elderly patient with noninfectious uveitis who failed to respond to anti-inflammatory therapy raises suspicion for PVRL. When the visual acuity does not correlate and is much better than the findings of numerous cells in sheets, clumps, and haze in the vitreous (Fig. 1), the discrepancy may also suggest the presence of PVRL. The characteristic fundoscopic appearance of PVRL is yellowish orange subretinal infiltrates (Fig. 3) that may enlarge and coalesce over a short time.

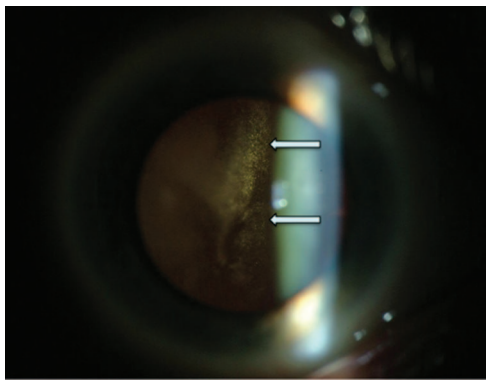

Figure 1. Slit-lamp biomicroscopy of a patient with primary vitreoretinal lymphoma. Binocular slit-lamp examination reveals numerous infiltrating cells (arrows) behind the lens in the vitreous.

Ocular imaging can be helpful for the diagnosis. Fundus autofluorescence may demonstrate multiple weak or bright hyperfluorescent spots (Fig. 4), which suggest the overlapping of PVRL and RPE cells, and hypofluorescent areas, which suggest PVRL cells above the RPE cells or RPE atrophy [56]. Fluorescein angiography often shows clusters of round, hyper- or hypofluorescent lesions, $\sim$100 μm in size (Fig. 4), at the early and late phases in the posterior pole and heterogeneous choroidal fluorescence [54]. RPE disturbances are characterized by granularity, blockage, and late staining [57]. OCT images demonstrate hyper-reflective nodules at the RPE level (Fig. 2) [54], which likely correspond to subretinal PVRL cells between the RPE and Bruch's membrane. Because PVRL is closely related to PCNSL, it is essential to evaluate the CNS, including contrast-enhanced cranial magnetic resonance imaging (MRI) and CSF evaluation. The findings of lymphoma cells in CSF and CNS lesion(s) support the diagnosis of PVRL and may avoid a diagnostic procedure to obtain and examine ocular tissues.

Although physical examination and ocular imaging are useful adjuncts, the gold standard for PVRL diagnosis is the identification of PVRL cells in the eye. This requires surgical intervention, including aqueous aspiration, diagnostic vitrectomy, diagnostic retinal or chorioretinal biopsy, and, rarely, diagnostic enucleation [58]. Cytology and histopathology provide morphological evidence of PVRL, which is characterized by large, atypical lymphoid cells with large, irregular nuclei, prominent nucleoli, and scanty basophilic cytoplasm (Fig. 5A) [59]. Mitoses may be present, rare, or absent. Reactive lymphocytes are often mixed among the PVRL cells. PVRL cells easily undergo necrosis, so careful and prompt processing of the specimen (especially ocular fluids) is critical. Monoclonality, either a B-cell population with κ or λ restriction or a

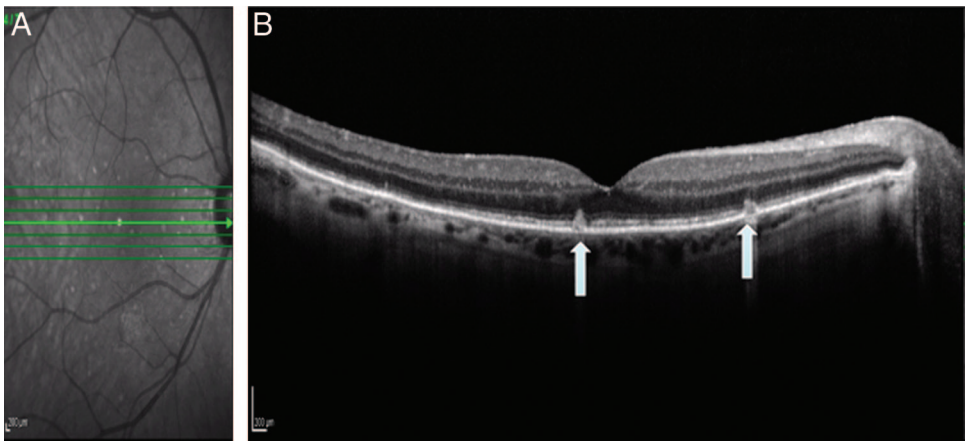

Figure 2. Optical coherent tomography (OCT) of the patient in Figure 1. **(A):** Cross-sectional images of the retina through several bright lesions are examined. **(B):** OCT shows nodular hyperreflective lesions (arrows) at the retinal pigment epithelium (RPE) level and sub-RPE space.

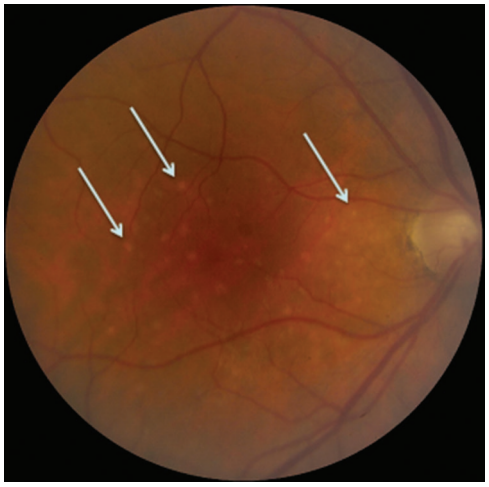

Figure 3. Fundoscopy of the patient in Figure 1. There are many small, round, yellow-orange lesions (arrows) at the retinal pigment epithelium level in the deep retina.

T-cell population, is observed using immunohistochemistry or flow cytometry. Immunohistochemistry is helpful in showing $CD20^+$ cells (Fig. 5B). In general, B cells are rarely part of nonmalignant autoimmune uveitis. There can be reactive $CD3^+$ T cells as well, and sometimes these outnumber the malignant $CD20^+$ cells, which may be misread as evidence for an inflammatory process.

Elevation of IL-10 levels in the ocular fluid and/or an IL-10:IL-6 ratio >1 is highly suggestive of, but not diagnostic of, B-cell PVRL [60–63], although T-cell PVRL might be associated with higher IL-10 levels in the vitreous [5]. Aqueous IL-10 levels are reported to correlate with clinical response to local chemotherapy and might be used as a biomarker for PVRL. The assumption of IL-10 as the sole biomarker for the diagnosis of PVRL should not be held if there is no other evidence of disease. Molecular analysis with microdissection and polymerase chain reaction is used to detect Ig_H gene rearrangements in B-cell lymphoma and T-cell receptor gene rearrangements in T-cell lymphoma [64]. The limitations of molecular testing, particularly with the small samples available from the eye, can give a false-positive result or a false-negative result. Therefore, microdissection identifying the atypical lymphoid cells is critical for molecular analysis. This highlights the importance of histology. These two immunological and molecular tests are useful adjuncts in diagnosing PVRL [55]. Currently, histology in combination with immunohistochemistry is probably most often used to make the diagnosis of PVRL.

Treatment

The optimal therapy for PVRL is not defined. Grimm et al. [25] reported, from an IPCG series of 221 immunocompetent patients with PCNSL and/or PVRL, that there was no difference with respect to disease progression or OS with the use of local versus systemic therapies. However, it is important to keep in mind that that study was an uncontrolled, multicenter, retrospective study involving many different treatments. Thus, it was not conclusively demonstrated whether or not ocular therapy has an impact on survival. At this time, some experts suggest reserving systemic therapy for those with CNS disease and using local therapy for disease confined to the eye; careful follow-up is indicated, and if there is evidence of CNS disease then treatment with systemic therapy should be used.

Local therapies include ocular radiation and intravitreal chemotherapy [65]. There has been no study to compare these treatment options, but there does not appear to be a significant difference with respect to local tumor control or treatment-related visual compromise. At the present time, some experts prefer intravitreal chemotherapy whereas others prefer ocular radiation as first-line therapy [66]. Until studies are done to adequately address this question, the treatment decision should probably be based on patient considerations, such as disease laterality, the distance the patient must travel for treatment, and patient preference. In general, intravitreal chemotherapy is only injected into the eye with PVRL lesions.

External-beam radiotherapy (EBRT) typically involves a total of 35–40 Gy delivered in ~15 interrupted fractions (2 Gy

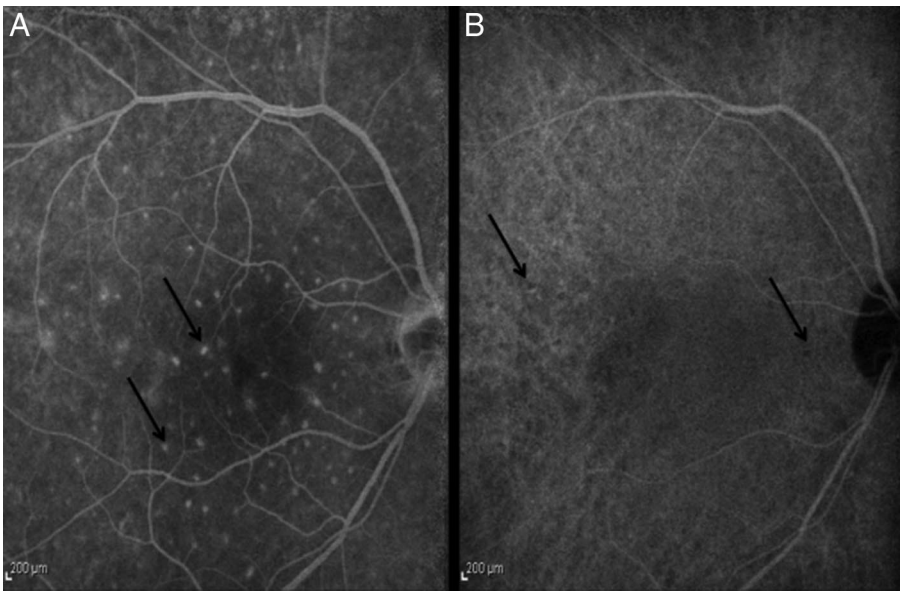

Figure 4. Fluorescein angiography and autofluorescence image of the patient in Figure 1. **(A):** There are small, round, hyperflorescent spots (arrows) corresponding to the lesions seen in Figure 3. **(B):** Fundus autofluorescence (fluorescence from the eye occurs without injection of dye) image shows small, round, hypofluorescent spots (arrows) reflecting lymphoma cells at the retinal pigment epithelium level.

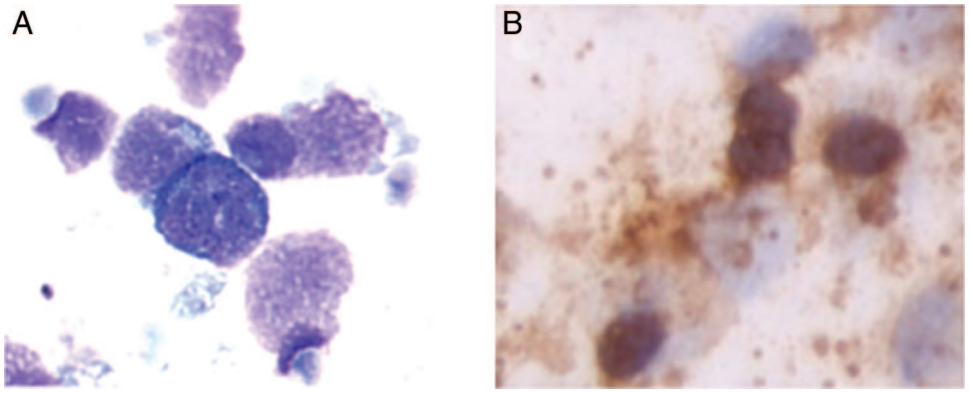

Figure 5. Cytology of primary vitreoretinal lymphoma. **(A):** Typical lymphoma cells are characterized by large nuclei, prominent nucleoli, and scanty basophilic cytoplasm (Giemsa stain; original magnification, ×640). **(B):** Immunohistochemistry illustrates $CD20^{+}$ cells that are brown in color (avidin-biotin-complex immunoperoxidase; original magnification, ×400).

per fraction) using opposed lateral beams to include the entirety of both eyes in the treatment ports. Using this technique, the rates of local recurrence and visually significant radiation retinopathy have been reported to be very low [42, 67, 68]. It is not necessary to attempt to spare the lens, which can be a cause of treatment failure by leaving untreated lymphoma cells in the anterior vitreous, because cataracts are easily managed using standard surgical techniques. EBRT may be preferable in patients with bilateral disease, whereas intravitreal chemotherapy (see below) may be preferred in patients with unilateral disease or in patients with previous ocular radiation. If EBRT is to be used in a patient with unilateral disease, both eyes should be treated because PVRL almost always becomes bilateral.

Intravitreal methotrexate was shown to be efficacious in small nonrandomized trials [23, 69–73]. Frenkel et al. [23] reported on the largest series of 44 eyes of 26 patients with PVRL. They demonstrated clinical remission after a mean of 6.4 ± 3.4 (range, 2–16) injections of methotrexate. Intravitreal rituximab was shown to penetrate the entire retina [24]. The half-life is approximately 5 days, so an intravitreal injection probably has efficacy for approximately 3–4 weeks [74]. Small, nonrandomized studies have demonstrated the activity of intravitreal rituximab monotherapy for PVRL [75, 76]. There is also some evidence that the combination of intravitreal methotrexate and rituximab may be effective for PVRL, although the results are preliminary and based on small patient numbers [77]. This alternating, combination approach is potentially attractive because it decreases the need for multiple methotrexate injections with concomitant toxicity. Regardless of which local therapy is used, there is a high likelihood of local recurrence and CNS involvement. The detection of early,

isolated ocular recurrence is difficult. Given the high risk for subsequent CNS involvement, it would seem reasonable to perform intermittent MRI of the brain. Notably, of the 56% of the 47 patients who relapsed, 62% had CNS lesions [24].

Because PVRLs are often high grade and can be associated with PCNSL in ~20% of cases, systemic chemotherapy has been investigated as a treatment modality, often with regimens that are used for the therapy of PCNSL. The reported systemic chemotherapy options can be divided into single-agent chemotherapy, combination chemotherapy, and high-dose chemotherapy plus autologous stem cell transplantation (ASCT).

An initial case report by Baumann et al. [78] reviewed the treatment of a 59-year-old female patient originally diagnosed with DLBCL of the breast who had intraocular relapse 1 year after primary therapy. She eventually achieved a complete remission (CR) for >1 year after treatment with high-dose arabinofuranosyl cytidine (Ara-C) at 2–3 g/m^2. A second report on six patients treated with high-dose Ara-C at 2–3 g/m^2 given at two different intervals demonstrated responses in seven of eight patients (one CR, six partial remissions [PRs]) [79]. The patient achieving a CR eventually relapsed, but was able to regain the CR after a second administration of high-dose Ara-C, which was then given as a monthly maintenance infusion. All the patients who achieved only a PR received radiation therapy as consolidation; therefore, the duration of remission from single-agent Ara-C could not be determined. Because of the efficacy of high-dose methotrexate in PCNSL patients, Batchelor et al. [27] reported on nine patients with intraocular involvement of lymphoma treated with methotrexate at 8 g/m^2. Potentially cytotoxic, micromolar levels of methotrexate were detectable in the aqueous and vitreous humor in most patients. Of the nine patients, two had PVRL alone, five had PVRL and PCNSL, and two had PVRL, CNS, and systemic disease as well. An intraocular response was reported in seven patients, with CRs in six and a PR in one. All seven patients with CNS involvement had resolution of their CNS disease. Relapse occurred in three of the seven responders. In a recent, prospective, single-center study by Jahnke et al. [80], 10 patients with PVRL were treated with ifosfamide or trofosfamide. There was a 100% response rate (nine CRs, one PR). The median PFS interval was 18 months, with a median OS duration of 32 months. Of the seven relapses, five were ocular and two were CNS. Thus, this class of agents plus high-dose Ara-C appears to have good penetration into the intraocular space and should be investigated further alone or in combination in the treatment of PVRL patients.

Combination chemotherapy trials in PVRL patients are extremely limited because the majority of studies have incorporated radiation into the treatment regimen and cannot provide an accurate assessment of the chemotherapeutic effect alone. Sandor et al. [81] reported on 14 patients, including five with intraocular involvement, treated with a complex treatment schema incorporating i.v. as well as i.t. chemotherapy without irradiation. Patients received i.v. methotrexate, vincristine, and thiotepa as well as i.t. methotrexate and Ara-C in 21-day cycles. There was a 100% response rate (11 CRs, three PRs). At a median of 4.5 years of follow-up, the PFS rate was 34.3% and the OS rate was 68.8%. Thus, although a high initial response was seen, the duration was limited and additional therapy is likely needed. Most recently, the use of methotrexate, rituximab, and temozolomide was advocated for the therapy of PCNSL patients, but studies are still ongoing.

Several groups have reported their experience with high-dose chemotherapy and ASCT. Soussain et al. [82] reported on 11 patients with PVRL who were treated with etoposide, methylprednisolone, high-dose Ara-C, and cisplatin and/or high-dose methotrexate. First-line therapy failed in 10 of 11 patients. Five of the refractory patients then underwent salvage chemotherapy with thiotepa, busulfan, and cyclophosphamide followed by high-dose chemotherapy and ASCT, and all achieved a CR. Unfortunately, two patients relapsed at a median of 6 months and three remained alive and in remission at a median follow-up of 14 months. Unpublished data from the Mayo Clinic (Rochester, MN) identified six patients (five with PVRL and one with PCNSL with ocular involvement) who had undergone ASCT following BCNU, etoposide, cytarabine, and melphalan conditioning. With a median follow-up of 3 years, 50% of patients remained in CR with no additional therapy. In a multicenter, phase II clinical trial, 43 patients with relapsed or refractory PCNSL, including 10 with intraocular involvement, were treated with salvage chemotherapy and 27 subsequently underwent high-dose chemotherapy with ASCT [83]. Patients who underwent ASCT had longer PFS and OS times than those who did not, and on univariate analysis it did not appear that patients with intraocular involvement did worse than those without eye involvement. In summary, although systemic chemotherapy alone can result in high response rates in patients with PVRL, there is an extremely high relapse rate. Systemic chemotherapy probably should be initiated only once there is evidence of PCNSL or systemic lymphoma.

Treatment toxicities depend on the therapeutic modality. For local therapy, toxicities include complications of intraocular injections seen with monthly injections of anti–vascular endothelial growth factor for neovascular age-related macular degeneration, the overall incidence of which is quite low [84–86]. Complications of intravitreal injection include cataracts, vitreous hemorrhage, endophthalmitis, and retinal detachment [87]. A specific toxicity of intravitreal methotrexate is epithelial keratopathy [23, 72], which can be reduced by a paracentesis before the injection [88]. Also, cataracts and hypotony may develop; the latter is noted only after about 12 injections. Intravitreal rituximab rarely causes vitritis. Local irradiation can cause radiation retinopathy, especially if there are other predisposing risk factors such as diabetes mellitus or hypertension. The dose that might cause the development of radiation retinopathy is under contention, but the risk in the absence of other risk factors for the development of retinopathy is probably low up to 35 Gy but increases after that point [89–91].

For systemic therapy, there is a risk for giving excessive frontal lobe radiation if local ocular radiation has been given and, in the future, the patient receives whole-brain radiotherapy. The converse is also a consideration. If there is concern that the patient might need whole-brain radiotherapy in the fu-

ture, the use of local EBRT to the eye should be avoided. The toxicities seen with systemic therapy are inherent to each individual chemotherapeutic agent; however, in general, myelosuppression is the most commonly seen toxicity. Combination chemotherapy, especially when combined with stem cell transplantation, can result in neutropenia, increasing the risk for febrile neutropenia and potential mortality. Thus, vigilance must be maintained in patients receiving systemic chemotherapy.

Notwithstanding, at this time we recommend the following guidelines to treat PVRL patients who do not enroll in a clinical trial:

1. Without CNS or systemic involvement: (a) If only one eye involved, use local therapy. Whether it is local intravitreal methotrexate and rituximab given alone or carefully given between 30–35 Gy of EBRT is still under contention. (b) If both eyes involved, there is still a preference toward local therapy, although systemic treatment should not be excluded. The addition of intravitreal chemotherapeutic agents to systemic therapy should be considered.

2. With CNS involvement: (a) High-dose methotrexate-based therapy (possibly with systemic rituximab) is recommended in conjunction with local therapy given the limited penetration of systemic agents into the vitreous cavity. (b) Whole brain radiotherapy in conjunction with ocular radiotherapy should be considered in those who have failed systemic therapy and are too debilitated or do not meet the criteria for more aggressive therapy such as ASCT.

The National Comprehensive Cancer Network has also proposed an alternative but similar recommendation under the treatment guidelines for CNS cancers [92].

Summary and Future Studies

PVRL commonly masquerades as posterior uveitis and is often a fatal disease because of a close association with PCNSL. An accurate diagnosis is required to treat the patient. Oncologists, ophthalmologists, and pathologists should work together to investigate the pathogenesis and improve methods for diagnosis and treatment of this malignancy. Because of the disease rarity, this requires international, multicenter, collaborative efforts. Priorities for future studies include: the application of transcriptional and proteomic profiling studies to ocular pathological specimens to elucidate mechanisms, the development of new treatment approaches using animal models, clinical trials to evaluate and compare existing therapeutic options, and investigation of new ophthalmic imaging modalities, such as optical coherence tomography and intraocular IL-10 levels (or the IL-10:IL-6 ratio) to follow treatment responses. Treatment recommendations outside the context of a clinical trial include chemotherapy and radiation therapies (local, systemic or combined), and are generally based on PVRL with or without CNS involvement.

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